

Attorney Docket No.: 44158/244344 (SJ-0029)  
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This listing of the claims will replace all prior versions and listings of claims in the application:

Listing of the claims:

Claim 1: (currently amended) A method for predicting the ~~level and distribution of~~ CYP3A5 expression level in a subject comprising determining the nucleotide present in each CYP3A5 allele of the genomic DNA of said subject at the location(s) selected from the group consisting of:

(a) the position corresponding to nucleotide ~~22,893~~ of ~~Genbank sequence accession no. AC005020~~ 23 of SEQ ID NO:73 within intron 3 of the Cyp3A5 gene:

(b) the position corresponding to nucleotide ~~30,597~~ of ~~Genbank sequence accession no. AC005020~~ 29 of SEQ ID NO:74 within exon 7 of the Cyp3A5 gene: and

(c) the positions corresponding to both nucleotide ~~22,893~~ and nucleotide ~~30,597~~ of ~~Genbank sequence accession no. AC005020~~ 23 of SEQ ID NO:73 and nucleotide 29 of SEQ ID NO:74;

wherein the presence of an A at the position corresponding to nucleotide ~~22,893~~ of ~~Genbank sequence accession no. AC005020~~ 23 of SEQ ID NO:73 on at least one CYP3A5 allele of said subject predicts a relatively high level of expression of CYP3A5 and the

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presence of a U at the position corresponding to nucleotide ~~22,893 of Genbank sequence accession no. AC005020~~ 23 of SEQ ID NO:73 on each CYP3A5 allele of said subject predicts a relatively low level of expression;

wherein the presence of a G at the position corresponding to nucleotide ~~30,597 of Genbank sequence accession no. AC005020~~ 29 of SEQ ID NO:74 on at least one CYP3A5 allele of said subject predicts a relatively high level of expression of CYP3A5 and the presence of an A at the position corresponding to nucleotide ~~30,597 of Genbank sequence accession no. AC005020~~ 29 of SEQ ID NO:74 on each CYP3A5 allele of said subject predicts a relatively low level of expression of CYP3A5; and wherein the presence of an A at the position corresponding to nucleotide ~~22,893 of Genbank sequence accession no. AC005020~~ 23 of SEQ ID NO:73 and a G at the position corresponding to nucleotide ~~30,597 of Genbank sequence accession no. AC005020~~ 29 of SEQ ID NO:74 on at least one CYP3A5 allele of said subject predicts a relatively high level of expression of CYP3A5 and the presence of either a U at the position corresponding to nucleotide ~~22,893 of Genbank sequence accession no. AC005020~~ 23 of SEQ ID NO:73 or an A at the position corresponding to nucleotide ~~30,597 of Genbank sequence accession~~

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~~no. AC005020~~ 29 of SEQ ID NO:74 on each CYP3A5 allele of said subject predicts a relatively low level of expression of CYP3A5.

Claim 2: (currently amended) The method of claim 1 wherein said location is the position corresponding to nucleotide ~~22,893~~ of Genbank sequence accession no. AC005020 23 of SEQ ID NO:73 within intron 3 of the Cyp3A5 gene.

Claim 3: (currently amended) The method of claim 1 wherein said location is the position corresponding to nucleotide ~~30,597~~ of Genbank sequence accession no. AC005020 exon 5 29 of SEQ ID NO:74 within exon 7 of the Cyp3A5 gene.

Claim 4: (currently amended) The method of claim 1 wherein said locations are the positions corresponding to both nucleotide ~~22,893~~ and nucleotide ~~30597~~ of Genbank sequence accession no. AC005020 23 of SEQ ID NO:73 and nucleotide 29 of SEQ ID NO:74.

Claim 5: (currently amended) The method of claims ~~1-4~~ 1, 2, 3 or 4 wherein the step of determining the nucleotide present in each CYP3A5 allele of said subject at the selected location(s) is

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accomplished by sequencing a region of the genomic DNA of said subject which includes said location(s).

Claim 6: (currently amended) The method of claims ~~1-4~~ 1, 2, 3 or 4 wherein the step of determining the nucleotide present in each Cyp3A5 allele of said subject at the selected location(s) is accomplished by

(a) amplifying a region of the genomic DNA of said subject which includes said location(s) to generate an amplified fragment, and

(b) treating the amplified fragment with a restriction enzyme in its corresponding restriction buffer to determine the identity of the nucleotide present at the selected location(s).

Claim 7: (currently amended) The method of claims ~~1-4~~ 1, 2, 3 or 4 wherein the step of determining the nucleotide present in each Cyp3A5 allele of said subject at the selected location(s) is accomplished by

(a) amplifying a region of the genomic DNA of said subject which includes said location(s), and

(b) hybridizing the amplified region with probes specific

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for the selected location(s) wherein hybridization determines the identity of the nucleotide present at the selected location(s).

Claim 8: (currently amended) A method for determining the cytochrome P450 3A5 (CYP3A5) genotype and phenotype of an individual comprising:

- (a) isolating nucleic acid from the individual;
- (b) amplifying a region of the cytochrome P450 3A5 (CYP3A5) gene sequence selected from the group of:
  - (i) intron 3 comprising the position corresponding to nucleotide 22,893 of Genbank accession no. AC005020 23 of SEQ ID NO:73;
  - (ii) exon 7 comprising the position corresponding to nucleotide 30,597 of Genbank accession no. AC005020 29 of SEQ ID NO:74; and
  - (iii) intron 3 comprising the position corresponding to nucleotide 22,893 of Genbank accession no. AC005020 23 of SEQ ID NO:73 and exon 7 comprising the position corresponding to nucleotide 30,597 of Genbank accession no. AC005020 29 of SEQ ID NO:74; and
- (c) ~~analyzing the cytochrome P450 3A5 (CYP3A5) sequence~~

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sequencing the amplified region of step (b), thereby determining the cytochrome P450 3A5 (CYP3A5) genotype and phenotype of the individual.

Claim 9: (currently amended) The method of claim 8 wherein the intron 3 region of cytochrome P450 3A5 (CYP3A5) is amplified utilizing primers which amplify 5' and 3' of the nucleotide ~~22,893 of Genbank accession no. AC005020~~ position corresponding to nucleotide 23 of SEQ ID NO:73.

Claim 10: (currently amended) The method of claim 9 wherein the intron 3 region is amplified utilizing ~~primer pairs~~ SEQ ID NO: 24 and 25 primers, or ~~primer pairs~~ SEQ ID NO: 26 and 27 primers.

Claim 11: (currently amended) The method of claim 8 wherein the exon 7 region of cytochrome P450 3A5 (CYP3A5) is amplified utilizing primers which amplify 5' and 3' of the nucleotide ~~30,597 point mutation of Genbank accession no. AC005020~~ position corresponding to nucleotide 29 of SEQ ID NO:74.

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Claim 12: (currently amended) The method of claim 11 wherein the exon 7 region is amplified utilizing ~~primer pairs~~ SEQ ID NO: 30 and 16 primers, or ~~primer pairs~~ SEQ ID NO: 31 and 32 primers.

Claim 13: (currently amended) A method for determining cytochrome P450 3A5 (CYP3A5) intron 3 genotype of a subject which comprises:

(a) isolating nucleic acid from said subject;

(b) amplifying a cytochrome P450 3A5 (CYP3A5) PCR fragment from said nucleic acid using a set of primers, wherein said set of primers contains primer X and primer Y; wherein

(i) the X primer is complementary to a region 5' to the ~~point mutation site at nucleotide 22,893 of Genbank accession no. AC005020~~ position corresponding to nucleotide 23 of SEQ ID NO:73;  
and

~~(iii)~~ (ii) the Y primer is complementary to a region 3' to the ~~point mutation site at nucleotide 22,893 of Genbank accession no. AC005020~~ position corresponding to nucleotide 23 of SEQ ID NO:73;

~~(c) amplifying and the sequence cytochrome P450 3A5 (CYP3A5) PCR fragment amplified is in between primers X and Y, thereby~~

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~~obtaining an amplified fragment; and~~

~~(d)~~ (c) sequencing the amplified fragment obtained in step  
~~(e)~~ (b), thereby determining the cytochrome P450 3A5 (CYP3A5)  
intron 3 genotype of said subject.

Claim 14: (original) The method of claim 13 wherein primer X has the sequence corresponding to SEQ ID NO: 24, or a fragment thereof which is at least ten bases long, and primer Y has the sequence corresponding to SEQ ID NO: 25, or a fragment thereof which is at least ten bases long.

Claim 15: (original) The method of claim 13 wherein primer X has the sequence corresponding to SEQ ID NO: 26, or a fragment thereof which is at least ten bases long, and primer Y has the sequence corresponding to SEQ ID NO: 27, or a fragment thereof which is at least ten bases long.

Claim 16: (currently amended) A method for determining cytochrome P450 3A5 (CYP3A5) genotype of a subject which comprises:

(a) isolating nucleic acid from said subject;



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(b) making a first and a second PCR primer wherein

(i) the first PCR primer is complementary to intron 3 and introduces a base change in the PCR product adjacent to or near the ~~point mutation at nucleotide 22,893 of Genbank accession no. AC005020~~ position corresponding to nucleotide 23 of SEQ ID NO:73, such that a restriction site is generated in the presence of a particular nucleotide at the position corresponding to nucleotide 22,893 23 of SEQ ID NO:73 in the PCR product; and

(ii) the second PCR primer is complementary to a region 3' to the ~~intron 3 nucleotide 22,893 of Genbank accession no. AC005020~~ position corresponding to nucleotide 23 of SEQ ID NO:73 of intron 3;

(c) amplifying the sequence in between the first and the second primers; thereby obtaining an amplified fragment; and

(d) treating the amplified fragment obtained in step (c) with a restriction enzyme in its corresponding restriction buffer to detect presence or absence of a point mutation at ~~nucleotide 22,893 of Genbank accession no. AC005020~~ the position corresponding to nucleotide 23 of SEQ ID NO:73, thereby determining the cytochrome P450 3A5 (CYP3A5) genotype of said subject.

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Claim 17: (currently amended) The method of claim 16 wherein the first primer introduces a *Tru9I*/*MseI* restriction site in the presence of an A nucleotide at ~~nucleotide 22,893~~ the position corresponding to nucleotide 23 of SEQ ID NO:73, and the second primer has the sequence selected from SEQ ID NO:27 and SEQ ID NO:25, or a fragment thereof which is at least ten bases long.

Claim 18: (original) The method of claim 16 wherein the first primer has the sequence corresponding to SEQ ID NO: 33, or a fragment thereof which is at least ten bases long, and the second primer has the sequence corresponding to SEQ ID NO: 27, or a fragment thereof which is at least ten bases long.

Claim 19: (original) The method of claim 16 wherein the first primer has the sequence corresponding to SEQ ID NO:33 , or a fragment thereof which is at least ten bases long, and the second primer has the sequence corresponding to SEQ ID NO:25, or a fragment thereof which is at least ten bases long.

Claim 20: (currently amended) A method for determining cytochrome P450 3A5 (CYP3A5) genotype of a subject which

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comprises:

(a) isolating nucleic acid from said subject;

(b) amplifying a cytochrome P450 3A5 (CYP3A5) PCR fragment from said nucleic acid using a first set of primers, wherein said first set of primers contains primer X and primer Y; wherein

(i) the X primer is complementary to a region 5' to the ~~point mutation site at nucleotide 22,893 of Genbank accession no. AC005020~~ position corresponding to nucleotide 23 of SEO ID NO:73;  
and

(ii) the Y primer is complementary to a region 3' to the ~~point mutation site at nucleotide 22,893 of Genbank accession no. AC005020~~ position corresponding to nucleotide 23 of SEO ID NO:73;

~~(c) amplifying and the sequence cytochrome P450 3A5 (CYP3A5) PCR fragment amplified is~~ in between primers X and Y, thereby obtaining an first round amplified fragment;

~~(d) (c)~~ amplifying the first round amplified fragment of step (b) using a second set of primers, wherein said second set of primers contains primer Z and primer W, wherein

(i) primer Z is complementary to intron 3 and introduces a base change in the PCR product adjacent to or near

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~~the point mutation at nucleotide 22,893 of Genbank accession no. AC005020~~ position corresponding to nucleotide 23 of SEQ ID NO:73,  
such that a restriction site is generated in the presence of a  
particular mutation at ~~nucleotide 22,893~~ the position  
corresponding to nucleotide 23 of SEQ ID NO:73; and

(ii) primer W is complementary to a region 3' to intron  
3;

~~(e) amplifying and the amplified sequence is in between~~  
primers Z and W, ~~thereby obtaining an amplified fragment; and~~

~~(f) (d) treating the amplified fragment obtained in step (e)~~  
(c) with a restriction enzyme in its corresponding restriction  
buffer to detect presence or absence of a point mutation at  
~~nucleotide 22,893 of Genbank accession no. AC005020~~ the position  
corresponding to nucleotide 23 of SEQ ID NO:73, thereby  
determining the cytochrome P450 3A5 (CYP3A5) genotype of said  
subject.

Claim 21: (currently amended) The method of claim 20 wherein  
primer X has the sequence corresponding to SEQ ID NO: 24, or a  
fragment thereof which is at least ten bases long; primer Y has  
the sequence selected from the group of SEQ ID NO:25, or a

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fragment thereof which is at least ten bases long; primer Z introduces a *Tru9I/MseI* restriction site in the presence of an A nucleotide at ~~nucleotide 22,893 of Genbank accession no. AC005020~~ the position corresponding to nucleotide 23 of SEQ ID NO:73; and primer W has the sequence selected from SEQ ID NO: 27 and SEQ ID NO: 25, or a fragment thereof which is at least ten bases long.

Claim 22: (original) The method of claim 21 wherein primer Z has the sequence corresponding to SEQ ID NO: 33, or a fragment thereof which is at least ten bases long.

Claim 23: (currently amended) A method for determining cytochrome P450 3A5 (CYP3A5) genotype of a subject which comprises

- (a) isolating nucleic acid from said subject;
- (b) amplifying a cytochrome P450 3A5 (CYP3A5) PCR fragment from said nucleic acid using a set of primers, wherein said set of primers contains primer X and primer Y; wherein
  - (i) the X primer is complementary to a region 5' to the ~~point mutation site at nucleotide 30,597 of Genbank accession no. AC005020~~ position corresponding to nucleotide 29 of SEQ ID NO:74;

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~~(iii)~~ (ii) the Y primer is complementary to a region 3' to the ~~point mutation site at nucleotide 30,597 of Genbank accession no. AC005020~~ position corresponding to nucleotide 29 of SEQ ID NO:74;

~~(c) amplifying and the sequence amplified cytochrome P450 3A5 (CYP3A5) PCR fragment is in between primers X and Y, thereby obtaining an amplified fragment; and~~

~~(d) (c)~~ sequencing the amplified fragment obtained in step ~~(c)~~ (b), thereby determining the cytochrome P450 3A5 (CYP3A5) exon 7 genotype of said subject.

Claim 24: (original) The method of claim 23 wherein primer X has the sequence corresponding to SEQ ID NO: 30, or a fragment thereof which is at least ten bases long, and primer Y 20 has the sequence corresponding to SEQ ID NO: 16, or a fragment thereof which is a least ten bases long.

Claim 25: (original) The method of claim 23 wherein primer X has the sequence corresponding to SEQ ID NO:31, or a fragment thereof which is at least ten bases long, and primer Y 25 has the sequence corresponding to SEQ ID NO:32 or a fragment thereof

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which is at least ten bases long.

Claim 26: (currently amended) A method for determining cytochrome P450 3A5 (CYP3A5) genotype of a subject which comprises:

- (a) isolating nucleic acid from said subject;
- (b) making a first and a second PCR primer wherein

(i) the first PCR primer is complementary to exon 7 and introduces a base change in the PCR product adjacent to or near the ~~point mutation at nucleotide 30,597 of Genbank accession no. AC005020~~ position corresponding to nucleotide 29 of SEQ ID NO:74, such that a restriction site is generated in the presence of a particular nucleotide at ~~nucleotide 30,597~~ the position corresponding to nucleotide 29 of SEQ ID NO:74; and

(ii) the second PCR primer is complementary to a region 3' to the ~~intron 3~~ exon 7 nucleotide ~~30,597 of Genbank accession no. AC005020~~ in the position corresponding to nucleotide 29 of SEQ ID NO:74;

(c) amplifying the sequence in between the first and the second primers; thereby obtaining an amplified fragment; and

(d) treating the amplified fragment obtained in step (c)

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with a restriction enzyme in its corresponding restriction buffer to detect presence or absence of a point mutation at ~~nucleotide 30,597 of Genbank accession no. AC005020~~ the position corresponding to nucleotide 29 of SEQ ID NO:74, thereby determining the cytochrome P450 3A5 (CYP3A5) genotype of said subject.

Claim 27: (currently amended) The method of claim 26 wherein the first primer introduces a *Tru9I/MseI* restriction site in the presence of a an A nucleotide at ~~nucleotide 30,597 of Genbank accession no. AC005020~~ the position corresponding to nucleotide 29 of SEQ ID NO:74, and the second primer has the sequence selected from SEQ ID NO:32 and SEQ ID NO:16, or a fragment thereof which is at least ten bases long.

Claim 28: (original) The method of claim 26 wherein the first primer has the sequence corresponding to SEQ ID NO: 34, or a fragment thereof which is at least ten bases long, and the second primer has the sequence corresponding to SEQ ID NO: 32, or a fragment thereof which is at least ten bases long.



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Claim 29 (original) The method of claim 26 wherein the first primer has the sequence corresponding to SEQ ID NO:34, or a fragment thereof which is at least ten bases long, and second primer has the sequence corresponding to SEQ ID NO:16, or a fragment thereof which is at least ten bases long.

Claim 30: (currently amended) A method for determining cytochrome P450 3A5 (CYP3A5) exon 7 genotype of a subject which comprises:

- (a) isolating nucleic acid from said subject;
- (b) amplifying a cytochrome P450 3A5 (CYP3A5) PCR fragment from said nucleic acid using a first set of primers, wherein said first set of primers contains primer X and primer Y; wherein
  - (i) the X primer is complementary to a region 5' to the ~~point mutation site at nucleotide 30,597 of Genbank accession no. AC005020~~ position corresponding to nucleotide 29 of SEQ ID NO:74;  
and
  - (ii) the Y primer is complementary to a region 3' to the ~~point mutation site at nucleotide 30,597 of Genbank accession no. AC005020~~ position corresponding to nucleotide 29 of SEQ ID NO:74;

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~~(c) amplifying and the sequence amplified cytochrome P450 3A5 (CYP3A5) PCR fragment is in between primers X and Y, thereby obtaining an a first round amplified fragment;~~

~~(d) (c) amplifying the first round amplified fragment of step (b) using a second set of primers, wherein said second set of primers contains primer Z and primer W, wherein~~

~~(i) primer Z is complementary to exon 7 and introduces a base change in the a PCR product adjacent to or near the point mutation at nucleotide 30,597 of Genbank accession no. AC005020 position corresponding to nucleotide 29 of SEO ID NO:74, such that a restriction site is generated in the presence of a particular mutation at nucleotide 30,597 of Genbank accession no. AC005020 the position corresponding to nucleotide 29 of SEO ID NO:74; and~~

~~(ii) primer W is complementary to a region 3' to exon 7;~~

~~(e) amplifying and the amplified sequence is in between primers Z and W, thereby obtaining an amplified fragment; and~~

~~(f) (d) treating the amplified fragment obtained in step (e) (c) with a restriction enzyme in its corresponding restriction buffer to detect presence or absence of a point mutation at~~

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~~nucleotide 30,597 of Genbank accession no. AC005020 the position~~  
~~corresponding to nucleotide 29 of SEQ ID NO:74,~~ thereby  
determining the cytochrome P450 3A5 (CYP3A5) genotype of said  
subject.

Claim 31: (currently amended) The method of claim 30 wherein  
primer X has the sequence corresponding to SEQ ID NO:30, or a  
fragment thereof which is at least ten bases long; primer Y has  
the sequence of SEQ ID NO: 16, or a fragment thereof which is at  
least ten bases long; primer Z introduces a *Tru9I/MseI*  
restriction site in the presence of an A nucleotide at ~~nucleotide~~  
~~30,597 the position corresponding to nucleotide 29 of SEQ ID~~  
~~NO:74;~~ and primer W has the sequence selected from SEQ ID NO:32  
and SEQ ID NO:16, or a fragment thereof which is at least ten  
bases long.

Claim 32: (original) The method of claim 31 wherein primer Z  
has the sequence corresponding to SEQ ID NO:34, or a fragment  
thereof which is at least ten bases long.

Claim 33-38: (canceled)

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